Microbiological Reviews

A Publication of the American Society for Microbiology

VOLUME 51 • DECEMBER 1987 • NUMBER 4

CONTENTS/SUMMARIES

Summary: Incompatibility is a phenomenon that everyone who works with plasmids has encountered and, one suspects, would like to understand in depth; it is ubiquitous, and its analysis has revealed fundamental concepts in the basic biology of plasmids and other replicons. According to the Jacob-Cuzin replicon hypothesis, incompatibility was initially thought to be due to competition for membrane attachment sites essential for replication and partitioning. However, its expression by integrated plasmids and by multicopy plasmids could not be reconciled with this view; a step in the right direction was Pritchard's hypothesis of autogenous negative replication control, which predicted that trans*-acting replication inhibitiors would be responsible; this prediction has been verified. However, it is not the whole story; although expressed by cloned fragments encoding such inhibitors, incompatibility, especially with multicopy plasmids, usually cannot be explained as simple inhibition of replication. Also, other elements of the plasmid replicon such as the replication origin and the partitioning system may also express incompatibility. In other words, incompatibility results from interference with any of the basic replicon maintenance functions. While it is easy to visualize the cloned inhibitor as an incompatibility determinant, it is less obvious why two isologous replicons must always segregate, whatever their copy numbers. In this review, the various types of incompatibility are summarized and their molecular mechanisms are explained insofar as possible. In considering these various mechanisms, we are led to the general hypothesis that incompatibility and certain other cases of plasmid instability are often due to interference with the ability of the target plasmid to correct fluctuations in copy number that arise inevitably in individual cells as a consequence of the random mode of plasmid replication. Inability to correct these fluctuations efficiently leads to a broadening of the frequency distribution of plasmid copies in individual cells, which results, in turn, in the appearance of plasmid negative segregants.*

Summary: Among the eucaryotic microorganisms, the cellular slime mold Dictyostelium discoideum is a favorite for research on transmembrane signal transduction. Physiologic studies with D. discoideum in the 1970s were followed by biochemical studies, defining the most prominent components of the system. Recent results suggest important homologies with signal transduction systems in vertebrates. The pharmacology and binding kinetics of the cell surface receptors for folic acid and cyclic adenosine

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3',5'-phosphate (cAMP) have been well defined, and the cAMP receptor has been isolated. Both receptors are probably coupled to G-proteins, regulating adenylate cyclase, a phosphatidyl inositol cycle, and guanylate cyclase. Receptor-mediated activation of guanylate cyclase in D. discoideum may occur via the phosphatidylinositol pathway. Desensitization to cAMP is associated with alteration in binding properties of the cAMP receptor and is accompanied by phosphorylation of the receptor protein. The present state of knowledge provides a starting point for the isolation of the presumed components of the system and their reconstitution into functional entities.

Summary: The process of adenovirus gene expression has been the subject of intense study over the past 10 years, principally owing to its utility as a model for eucaryotic gene expression and control. In particular, the study of control of the early adenoviral genes has been especially informative and valuable. The organization and expression of the early viral genes have been thoroughly documented. The control of transcription initiation from the various early promoters is a key aspect of viral gene control. Central to this regulation is the role of the E1A gene in activating viral transcription. Many of the details of this process are now known, including the identification of cellular transcription factors that are the apparent targets for E1A action. In view of the role of E1A in the alteration of cellular growth control and oncogenesis, the action of E1A via cellular transcription factors, to both stimulate and repress transcription, is of clear importance. The various aspects of early adenovirus gene expression that have come to light in recent years are reviewed and discussed. In particular, the details of E1A control, possible molecular mechanisms for the stimulatory effects, and the potential significance with respect to oncogenesis are addressed.

Summary: A variety of systems have been described that complement replication of otherwise growth-restricted deoxyribonucleic acid (DNA) viruses in the presence of another genetically divergent virus. DNA viruses of animals provide useful model systems for dissecting the molecular mechanisms underlying viral gene function, replication, and pathogenesis. In vitro studies defining a variety of DNA virus-virus interactions have contributed substantially to our current knowledge of viral infection. This review summarizes complementary DNA virus-virus interactions that allow replication of growth-restricted viruses in the presence of genetically unrelated viruses. Investigations of complementary virus interactions at the molecular level have enhanced our understanding of viral gene functions. Productive replication of adeno-associated virus, a naturally defective parvovirus, is absolutely dependent on coinfection with a helper virus. Three groups of DNA viruses, adenoviruses, herpesviruses, and poxviruses, can act as helpers. The interaction between adenoviruses and the herpesviruses provides a model system for studying virus gene regulation. Studies on activation of herpes simplex virus from a latent state by human cytomegalovirus is leading to a more complete understanding of latency and activation. The most extensively characterized complementary interaction between two viruses is that between the adenoviruses and the papovaviruses. The defective adenovirus-simian virus 40 hybrid virus is dependent on adenovirus transcapsidation for replication. The precise function necessary for simian virus 40 capsid protein synthesis as well as complementation of adenovirus replication in simian cells has been targeted to 14 base pairs at 0.193 map unit on the simian virus 40 map. The subset of events that enable defective or inactive animal DNA viruses to multiply, following complementation by a genetically unrelated virus, under conditions that typically do not support viral replication are described in this review. Although substantial advances have been made in our understanding of viral gene functions, continued efforts are needed to understand fully the molecular mechanisms of viral disease.

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Summary: The aminoglycoside-aminocyclitol antibiotics, polycationic inhibitors of protein synthesis, include such well-known antibacterial agents as streptomycin and gentamicin. Their target, the bacterial ribosome, is reached by traversing the cell envelope, including the cytoplasmic membrane which is a formidable barrier to charged solutes. Understanding the mechanism of this transmembrane movement has been the objective of a large body of kinetic, bioenergetic, and physiological studies. The kinetic properties of this uptake system are unusual. Despite its capacity to accumulate aminoglycosides against a concentration gradient, the system lacks both the specificity and the saturability commonly associated with active transport. Uptake depends on both electron transfer through the membrane-bound respiratory chain and a threshold membrane electrochemical potential ($\Delta\psi$). The rate of uptake varies substantially with the physiological status of the bacterial culture. Growth rate, alteration in composition of the respiratory chain, and global regulatory mechanisms all effect uptake rates in complex and interrelated ways. Recent attention has focused on the molecular interactions that occur during the induction phase that follows the initial exposure of bacterial cells to aminoglycosides. Induction requires aminoglycoside-sensitive ribosomes. It has been suggested that transmembrane channels are formed from aberrant proteins, products of translational misreading. Whether or not this hypothesis is correct, the permeability properties of cytoplasmic membranes do change during the induction phase. In addition to its intrinsic interest, an understanding of the principles governing aminoglycoside uptake could have significant practical value for treatment of infectious disease by providing a rational basis for therapy and by suggesting the formulation of new chemical structures with improved uptake properties and greater antibacterial efficacy.

Summary: The mechanisms for regulating gene expression in the yeast Saccharomyces cerevisiae have provided models for understanding the regulation of eucaryotic gene expression. One of the best understood genetic regulatory circuits in yeast cells is responsible for regulating expression of the GAL genes, which encode proteins involved in utilization of the sugar galactose. The central components of this regulatory circuit are the GAL4 protein, which binds to deoxyribonucleic acid and activates transcription of the GAL genes, and the GAL80 protein, which binds to the GAL4 protein and prevents it from activating transcription. An inducer molecule, derived from galactose, is thought to prevent the GAL80 protein from inhibiting GAL4 protein, thereby causing induction of GAL gene expression. A separate regulatory pathway, termed catabolite repression, prevents GAL gene expression during growth on glucose and is superimposed upon this galactose-specific regulatory circuit. The role of each of the components of the GAL gene regulatory mechanism is described in detail in this review. Many features of the mechanism, including positive control mediated by a deoxyribonucleic acid-binding protein required for gene expression and negative control by a protein that acts posttranscriptionally to inhibit function of the positive factor, are common to several fungal gene regulatory circuits and are similar to certain regulatory mechanisms in higher eucaryotes.

Summary: Adenosine 5′-triphosphate synthase is one of the best-studied membrane-bound enzymes. It plays a crucial role in oxidative as well as photophosphorylation and is common to both procaryotic and eucaryotic organisms. Regardless of source, the enzyme is composed of two parts: a membrane-peripheral portion (F_1) and a mem-

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brane-integral portion (F_0). According to the chemiosmotic hypothesis, the enzyme utilizes the electrochemical gradient of protons built up by either respiration or light to synthesize ATP. To this end, protons have to be translocated from one side of the membrane to the other via F_0. Recent genetic, molecular genetic, and biochemical studies shed some light on how this might work. This article critically reviews these data with respect to methodology. In particular, this article deals with bacterial F_0 preparations, how they differ from those obtained from eucaryotes, and what we can learn from dissociation and reconstitution experiments about the role of the single subunits in F_0. The article concludes with a general discussion of current hypotheses on the mechanism of H^+ translocation, which also bears on other membrane systems such as bacteriorhodopsin and enzymes of the respiratory chain.

Summary: The mechanism of coupling energy to transport of solutes into bacteria is often deduced from the dependence of the initial rate of transport or the steady-state level of solute accumulation on the proton motive force (or one of its components) or on another energy-rich (metabolic) intermediate. These analyses usually neglect possible regulatory effects of the internal or external pH or both. An analysis of amino acid transport into Streptococcus lactis and Streptococcus cremoris has revealed at least three different mechanisms of energy coupling: (i) transport driven by the proton motive force, (ii) transport driven by phosphate-bond energy, and (iii) exchange with another solute. The activities of most of these transport systems are under control of the internal pH. The pK_a values for the internal pH dependencies of these transport systems are around 7 and thus close to the internal pH maintained in streptococci. The rates of uptake of a number of essential nutrients (catalyzed by the glutamate/glutamine, leucine/isoleucine/valine, phosphate, and leucyl-leucine transport systems) increase with the internal pH, becoming optimal at alkaline pH values (around pH 7.5). In contrast, the rates of uptake of some nonessential solutes (catalyzed by the alanine/glycine and serine/threonine transport systems) decrease with increasing internal pH; they are optimal at around pH 6. The activities of some transport systems are also controlled by the external pH. External pH can change: (i) transport activity due to a change in affinity (K_t) or maximal activity (V_{max}) of the carrier protein or both; (ii) the available substrate concentration as a result of changes in the relative concentration of protonated species. The type of interaction of protons, from either the inner or the outer bulk water phase, with the transport proteins can be differentiated into catalytic and allosteric pH effects. The physiological implications of the observed regulatory pH effects on solute transport are discussed.

Summary: A study of high-affinity, siderophore-mediated iron assimilation led to a molecular mechanism whereby a cell regulates the absorption of an essential element, in this case, iron. In contrast to the enterobactin siderophore system of Escherichia coli, which is organized in several transcriptional units spread over more than 20 kilobases of deoxyribonucleic acid, the aerobactin system borne on ColV plasmids of hospital isolates of E. coli is confined to about 8 kilobases of deoxyribonucleic acid. The regulatory, biosynthetic, and transport genes are organized in a single transcriptional unit driven by a major strong promoter. A chromosomal lesion mapping at 15.7 min, fur (ferric uptake regulation), codes for a trans-acting factor which regulates all high-affinity iron uptake systems in E. coli. Isolation of the 17-kilodalton Fur protein and study of its properties in vitro, including a footprinting analysis, afforded a sequence for the aerobactin operator. This is the site of binding of the Fur repressor, in dimer form, following activation of the protein by ferrous ion. No other cofactors were required. Comparison of the well-characterized aerobactin promoter with the 5' regions of several other iron-regulated genes and operons in E. coli suggests an "iron box" operator consensus sequence consisting of the 19-base pair palindrome 5'GATAATGATAAT CATTATC. It is concluded that the cytosol of E. coli contains a pool of free or loosely bound Fe(II) that is available for activation of both ferrous iron-requiring enzymes and the Fur protein.

Date of Issue: 4 December 1987